METHOD FOR GENERATING LIBRARIES OF ANTIBODY GENES
COMPRISING AMPLIFICATION OF DIVERSE ANTIBODY DNAs
AND METHODS FOR USING THESE LIBRARIES FOR
THE PRODUCTION OF DIVERSE ANTIGEN COMBINING MOLECULES
Wigler, et al.
Appl. No.: Unassigned   *Atty Docket: STRATAG.7C2DV2C1*

Construct libraries of genes encoding
Ig heavy chain and/or light chain
in E. coli vector

↓

Increase diversity of libraries via
random mutagenesis (optional)

↓

Transfect libraries into cultured
cells, where they are expressed

↓

Identify cultured cells
expressing Ab of desired specificity

↓

Isolate gene(s) encoding Ab of
desired specificity and express

Fig. 1

METHOD FOR GENERATING LIBRARIES OF ANTIBODY GENES
COMPRISING AMPLIFICATION OF DIVERSE ANTIBODY DNAs
AND METHODS FOR USING THESE LIBRARIES FOR
THE PRODUCTION OF DIVERSE ANTIGEN COMBINING MOLECULES
Wigler, et al.
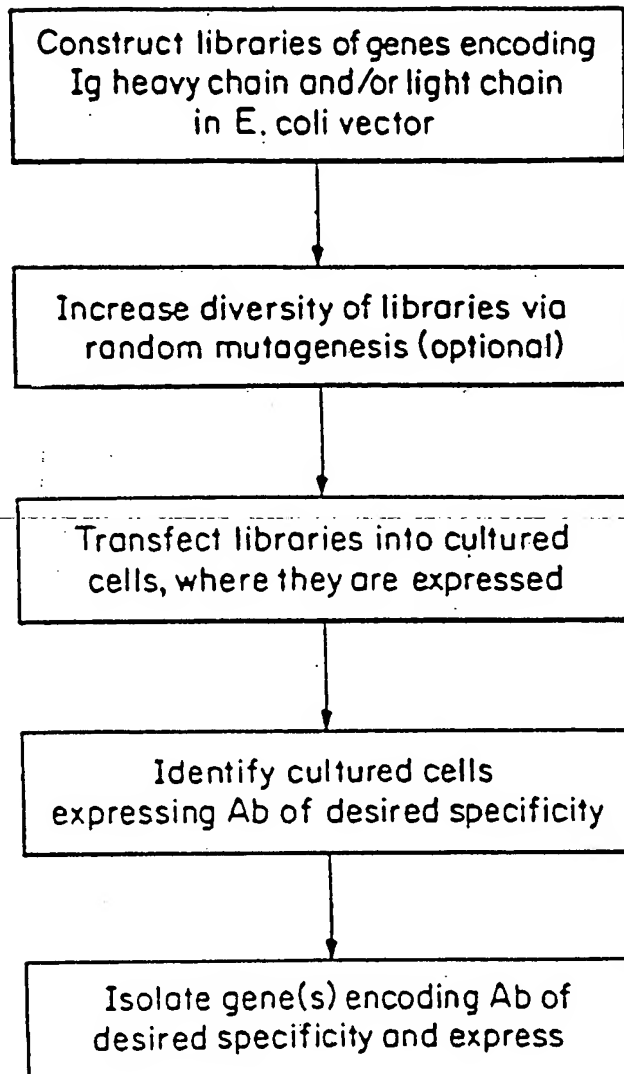*Appl. No.: Unassigned*   *Atty Docket: STRATAG.7C2DV2C1*

Fig. 2A

Fig. 2B

Fig. 2C

Fig. 2D

Fig. 2E

Fig. 2F

Fig. 2G

METHOD FOR GENERATING LIBRARIES OF ANTIBODY GENES
COMPRISING AMPLIFICATION OF DIVERSE ANTIBODY DNAs
AND METHODS FOR USING THESE LIBRARIES FOR
THE PRODUCTION OF DIVERSE ANTIGEN COMBINING MOLECULES
Wigler, et al.
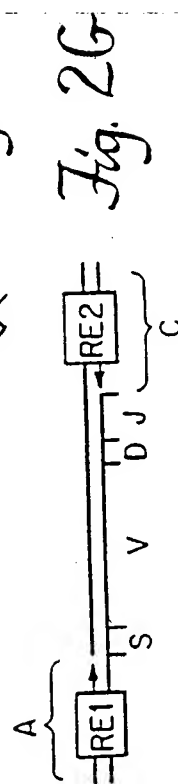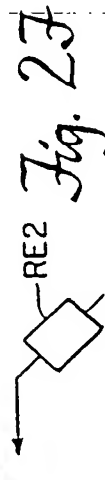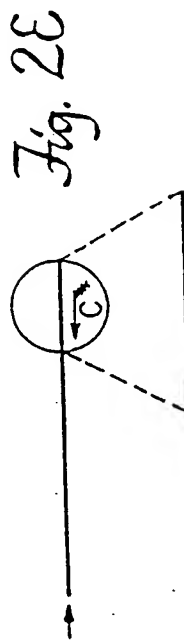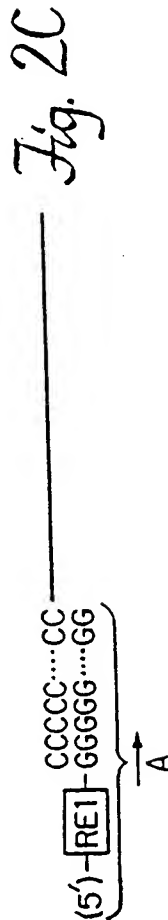Appl. No.: Unassigned   Atty Docket: STRATAG.7C2DV2C1

Fig. 3

**METHOD FOR GENERATING LIBRARIES OF ANTIBODY GENES
COMPRISING AMPLIFICATION OF DIVERSE ANTIBODY DNAs
AND METHODS FOR USING THESE LIBRARIES FOR
THE PRODUCTION OF DIVERSE ANTIGEN COMBINING MOLECULE**
Wigler, et al.
Appl. No.: Unassigned   *Atty Docket: STRATAG.7C2DV2C1*

```
          109             118             127
   >   AGT CAG TCC     TTC CCA AAT     GTC TTC ...
       Ser Gln Ser     Phe Pro Asn     Val Phe

          136             145             154
       GTC TCC TGC     GAG AGC CCC     CTG TCT GAT
       Val Ser Cys     Glu Ser Pro     Leu Ser Asp

          163             172             181
       AAG AAT CTG     GTG GCC ATG     GGC ...
       Lys Asn Leu     Val Ala MET     Gly

          190             199             208
       GCC CGG GAC     TTC CTG CCC     AGC ACC ATT
       Ala Arg Asp     Phe Leu Pro     Ser Thr Ile

          217             226             235
       TCC TTC ACC     GCC ... ...     AAC TAC CAG
       Ser Phe Thr     Ala             Asn Tyr Gln

          244             253             262
       AAC AAC ACT     GAA GTC ATC     CAG GGT AGA
       Asn Asn Thr     Glu Val Ile     Gln Gly Arg

          271             280             289
       ACC TTC CCA     AGG ACA TAC     AAC GGG ...
       Thr Phe Pro     Arg Thr Tyr     Asn Gly

          298             307             316
       GGC AAG CTA     GCC ATC TCG     CAG GTG TTG
       Gly Lys Leu     Ala Ile Ser     Gln Val Leu

          325             334             343
       CTG ACC AGC     TCT CCC AAG     AGC ...
       Leu Thr Ser     Ser Pro Lys     Ser

          352             361             370
       GGT TCA GAT     GAA TAC CTT     GAA ...
       Gly Ser Asp     Glu Tyr Leu     Glu

          379             388             397
       ... AAA ATC     CAC GTA TGC     AAC AGA GAT
           Lys Ile     His Val Cys     Asn Arg Asp

          406             415
       CTG CAT GTG     CCC ATT CCA  >  G
       Leu His Val     Pro Ile Pro
```
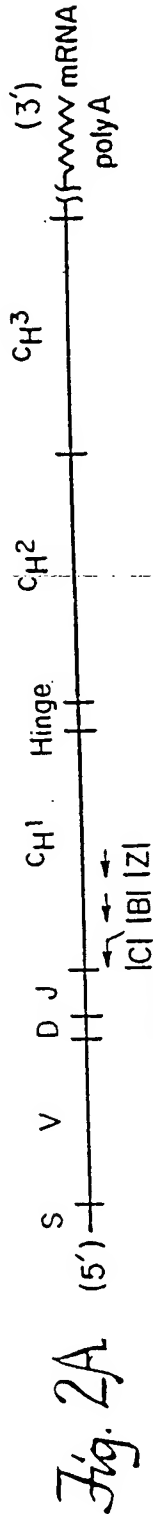
Fig. 4A

METHOD FOR GENERATING LIBRARIES OF ANTIBODY GENES
COMPRISING AMPLIFICATION OF DIVERSE ANTIBODY DNAs
AND METHODS FOR USING THESE LIBRARIES FOR
THE PRODUCTION OF DIVERSE ANTIGEN COMBINING MOLECULE

Wigler, et al.

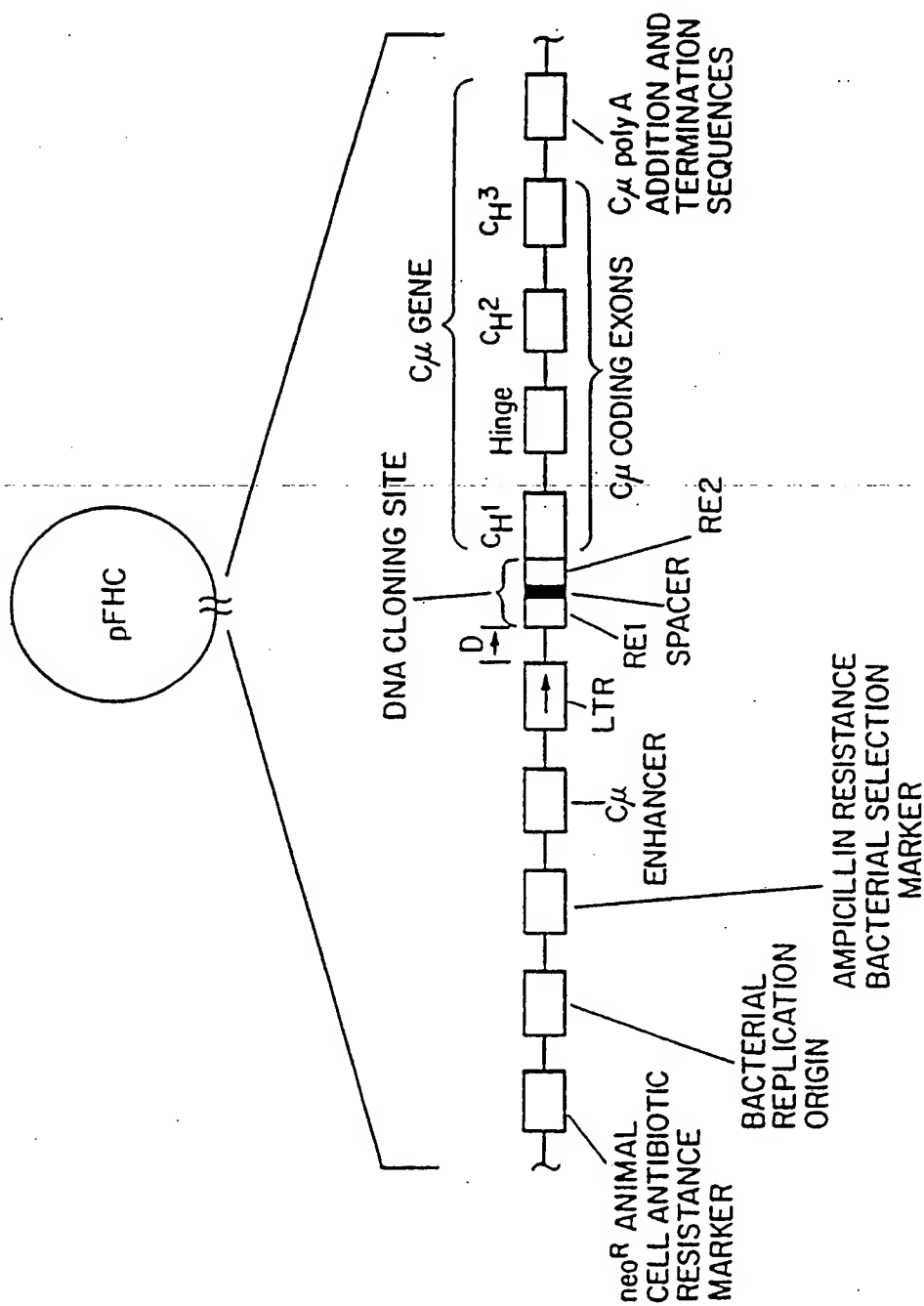Appl. No.: Unassigned   Atty Docket: STRATAG.7C2DV2CI

```
            109      118          127
        > AGN CAN TCN TTN CCN AAN GTN TTN
                  Ser         Pro     Val

136      145      154      163      172      181
GTN TCN  GAN AGN  CTN TCN  AAN AAN  CTN GTN  GCN
Val Ser       CCN Leu Ser       Leu Val  Gly
              Pro                 GCN ATN  GCN
                                  Ala      Gly

190      199      208      217      226      235
GCN GAN  CTN CCN  CTN CCN  ATN TCN  GTN GCN  CAN
Ala Arg  Leu      Leu Pro  Thr Ser  Val Ala
         TTN           ACN      TGN  AAN TAN
                       Thr      Thr

244      253      262      271      280      289
GAN GTN  CAN GGN  ATN AGN  TTN ACN  AGN ACN  GGN
    Val      Gly  Gly      Thr      Thr  Thr  Gly
                       ACN      CCN  AGN CTN
                       Thr      Pro      Leu

298      307      316      325      334      343
GCN      ACN TCN  GTN GTN  CTN TTN  CTN ATN  ACN
Leu Ala  Thr Ser  Val Val  Leu Ser  Leu      Thr
                            Pro ACN  ATN AGN
                                Thr      Leu

352      361      370      379      388      397
GAN TAN  CTN GTN  AAN ATN  TAN GGN  GGN AAN  GAN
Ser      Leu Val  Leu Val  Gly Gly  Gly Gly
TGN                CAN CAN  ATN CAN  AAN AGN  GAN
                        Pro           AAN

406      415
GTN CCN  > G
ATN CCN
Val Pro
Leu
Pro
```

Fig. 4B